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January 16, 2003

Mr. Phil Hutton
U.S. Environmental Protection Agency
Biopesticides and Pollution Prevention Division
Office of Pesticide Programs
Document Processing Desk (7504C)
Room 266A, Crystal Mall 2
1921 Jefferson Davis Highway
Arlington, VA 22202

Attention: Phil Hutton

AMEND SECTION G TO REVISE EXPERIMENTAL PROTOCOLS TO UPDATE EVENTS

BACILLUS THURINGIENSIS Cry34/35Ab1 INSECTICIDAL CRYSTAL PROTEINS AS EXPRESSED IN CORN (Construct PHP17658) (029964-EUP-U)

Dear Mr. Hutton:

Pioneer Hi-Bred International, Inc. is submitting an amended section G for the Experimental Use Permit application *BACILLUS THURINGIENSIS* Cry34/35Ab1 INSECTICIDAL CRYSTAL PROTEINS AS EXPRESSED IN CORN (Construct PHP17658) (029964-EUP-U) submitted on October 17, 2002.

The enclosed Section G: Proposed Experimental Program contains revised experimental protocols to reflect the current list of events that will be planted under this EUP (029964-EUP-U). The amended Section G has the confidentiality classification of 'A', except for the confidential appendix, which is classified as 'C.'

If you require further information, please contact me at 515-270-5983 or Larry Zeph, Registration Coordinator, at 515-253-5807.

Sincerely,

Isabelle S. Coats
Associate Registration Manager

Section G: Proposed Experimental Program

Experimental Use Permit Request
For
BACILLUS THURINGIENSIS CRY34/35Ab1 INSECTICIDAL CRYSTAL PROTEIN AS EXPRESSED IN MAIZE - CONSTRUCT PHP17658

January 16, 2003

Submitted By:

Pioneer Hi-Bred International, Inc.
7250 NW 62nd Avenue
Johnston, Iowa 50131

STATEMENT OF NO DATA CONFIDENTIALITY CLAIMS

No claim of confidentiality is made for any information contained in this study on the basis of its falling within the scope of FIFRA § 10(d) (1) (A), (B), or (C).

These data are the property of Pioneer Hi-Bred International, Inc. (Pioneer), and as such, are considered to be confidential for all purposes other than compliance with FIFRA §10. Submission of these data in compliance with FIFRA does not constitute a waiver of any right to confidentiality that may exist under any other statute or in any other country.

Company: Pioneer Hi-Bred International, Inc.

Company Agent: 

Isabelle S. Coats
Associate Registration Manager

Date: January 16, 2003

BACILLUS THURINGIENSIS CRY34/35Ab1 INSECTICIDAL CRYSTAL PROTEIN AS EXPRESSED IN MAIZE

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SECTION G: PROPOSED EXPERIMENTAL PROGRAM

Section G: Proposed Experimental Program

1. Participants

The experimental use program will be under the overall management of the following scientists:

Isabelle S. Coats, Ph.D.
Associate Registration Manager
Pioneer Hi-Bred Intenational, Inc.
7250 NW 62nd Avenue
Johnston, IA 50131

Paul Olson, Ph.D.
Research Manager-Disease Resistance
Pioneer Hi-Bred International, Inc.
7300 N. W. 62nd Avenue
Johnston, IA 50131

Laura Higgins, M.S.
Research Scientist - Insect Control/Herbicide Resistance
Pioneer Hi-Bred International, Inc.
7300 N. W. 62nd Avenue
Johnston, IA 50131

2. Target Pest and Overview of Experimental Program

The target pests to be evaluated in the proposed experimental program are ***Diabrotica virgifera virgifera*, Western corn rootworm (WCRW), and *Diabrotica berberi*, Northern corn rootworm (NCRW)**. Both insects are major pests of maize in North America. The development of transgenic maize expressing the binary ICP (insecticidal crystal protein) endotoxin will provide growers with a simple, highly effective, and environmentally benign means of controlling both rootworms. The transgenic maize tested under this EUP for plantings from March 2003 to March 2004 will be the first to use a CRY34/35Ab1 binary ICP derived from *Bacillus thuringiensis* strain PS149B1. Laboratory tests and small plot trials with transgenic CRY34/35Ab1 maize have shown that CRY34/35Ab1 has activity on corn rootworms (*Diabrotica* sp.). Further field testing across 19 states will continue to evaluate the efficacy of the CRY34/35Ab1 binary ICP. Additional objectives under the EUP experimental program include, conversion to commercial varieties, observation of agronomic potential, studies in resistant management and research into other commercial determinants. The proposed field protocols for these objectives are listed below in summary format. A complete description of each field protocol can be found on pages 7 through 24.

EUP Protocols	Acres	Lbs. of Seed
B.t. Cry34/35Ab1 Insect Resistance Management Trial	0.150	2.501
B.t. Cry34/35Ab1 Maize Agronomic Observation Trial	160.607	2,680.653
B.t. Cry34/35Ab1 Maize Breeding and Observation Nursery Trial	348.200	5,804.494
B.t. Cry34/35Ab1 Maize Demonstration Trial	5.202	86.717
B.t. Cry34/35Ab1 Maize Efficacy Trial	6.750	112.523
B.t. Cry34/35Ab1 Maize Hybrid Production Plots Trial	39.500	658.465
B.t. Cry34/35Ab1 Maize Regulatory Field Studies Trial	31.492	524.972
B.t. Cry34/35Ab1 Non-Target Trial	2.576	42.942
Herbicide Tolerance Study	27.750	462.593
Total	622.427	10,375.858

3. States and Acreage

PROPOSED EXPERIMENTAL USE PROGRAM
FOR PLANTINGS FROM MARCH 2003 THROUGH MARCH 2004

States	Maximum Transgenic Acres	Locations	Maximum Transgenic Seed	Max. Cry34Ab1 Protein in Seed Planted (grams) (32.5 ug per g dry weight)	Max. Cry35Ab1 Protein in Seed Planted (grams) (16.9 ug per g dry weight)
CA	2.875	2	71,875	0.706 g	0.037 g
GA	1.700	2	42,500	0.418 g	0.022 g
HW	339.850	8	8,496,250	83.501 g	4.342 g
IA	60.106	10	1,502,650	14.768 g	0.768 g
IL	46.656	11	1,166,400	11.463 g	0.596 g
IN	19.840	5	496,000	4.875 g	0.253 g
KS	7.236	6	180,900	1.778 g	0.092 g
MI	6.388	3	159,700	1.57 g	0.082 g
MN	21.072	7	526,800	5.177 g	0.269 g
MO	7.066	3	176,650	1.736 g	0.09 g
ND	6.850	3	171,250	1.683 g	0.088 g
NE	24.600	5	615,000	6.044 g	0.314 g
OH	3.280	6	82,000	0.806 g	0.042 g
PA	4.052	3	101,300	0.996 g	0.052 g
PW	45.150	6	1,128,750	11.093 g	0.577 g
SD	1.450	3	36,250	0.356 g	0.019 g
TN	1.950	2	48,750	0.479 g	0.025 g
TX	2.730	5	68,250	0.671 g	0.035 g
WI	19.576	7	489,400	4.81 g	0.25 g
TOTALS	622.427	97	15,560,675	152.93 g	7.952 g

B.t. CRY34/35Ab1 Insect Resistance Management Trial

Objective

These studies will provide information for the development of IRM strategies for genetically modified *B.t.* CRY34/35Ab1 maize lines expressing the binary ICP (insecticidal crystal protein). Specific experiments will focus on the determination of high dose against Western corn rootworm.

Description

Lines of each transformation event will be planted in up to four replications of a randomized design. Each line may be represented both by a segregate with the binary ICP and a segregate without the binary ICP. Experimental units will contain up to 3,000 plants. Plants will be artificially infested with Western corn rootworm or a natural infestation will be used. Data on the number of adults emerging from *B.t.* CRY34/35Ab1 plants versus non- *B.t.* CRY34/35Ab1 plants will be collected at various times during the growing season. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of locations
IA	2

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 0.050 acres per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings per site: up to two plantings per site.
Planting Dates: 3/1/03 - 8/1/04
Harvest Dates: 6/1/03 – 11/30/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue, whole plant samples, and/or insect samples may be taken up to ten times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed maybe returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35Ab1 Maize Agronomic Observation Trial

Objective

Assess agronomic and phenotypic effects of the binary ICP (insecticidal crystal protein) in genetically modified *B.t.* CRY34/35Ab1 maize lines.

Description

B.t. CRY34/35Ab1 maize lines will be observed for yield and other agronomic and phenotypic effects of insertion of the ICP. Plants may be treated with herbicide, and/or infested with Western corn rootworm or other corn insects, and/or sampled for various laboratory analyses to determine phenotype and segregation patterns. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test material will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of locations	State	Number of locations
CA	2	ND	3
GA	4	NE	5
HI	6	OH	2
IA	7	PA	2
IL	10	PR	6
IN	4	SD	2
KS	2	TN	2
MI	2	TX	2
MN	6	WI	6
MO	2		

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.00 acre per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings: Hawaii and Puerto Rico - up to 4 cycles with up to 5 plantings for a total of 20 plantings per year; all other states - up to 2 plantings per year.
Planting Dates: 3/1/03 - 3/1/04
Harvest Dates: 6/1/03 – 6/1/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue and/or whole plant samples may be taken several times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed may be returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35Ab1 Maize Breeding and Observation Nursery Trial

Objective

Through backcrossing, selfing, and observation of phenotype, convert non-genetically modified or genetically modified inbred lines to genetically modified *B.t.* CRY34/35Ab1 maize lines expressing the binary ICP (insecticidal crystal protein) and maintain these lines all the way to seed production.

Description

Using various experimental plot designs, *B.t.* CRY34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed. Plants may be treated with herbicide, and/or infested with Western corn rootworm or other corn insects, and/or sampled for various laboratory analyses to determine phenotype and segregation patterns. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of locations	State	Number of locations
HI	6	MN	3
IA	7	ND	2
IL	10	NE	2
IN	4	PR	6
KS	2		

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.00 acre per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings: Hawaii and Puerto Rico - up to 4 cycles with up to 5 plantings for a total of 20 plantings per year; all other states - up to 2 plantings per year.
Planting Dates: 3/1/03 - 3/1/04
Harvest Dates: 6/1/03 – 6/1/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue and/or whole plant samples may be taken several times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed may be returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35Ab1 Maize Demonstration Trial

Objective

Demonstrate the efficacy of genetically modified *B.t.* CRY34/35Ab1 expressing the binary ICP (insecticidal crystal protein) in controlling Western corn rootworm and/or other Coleopteran insects.

Description

Lines of each transformation event of *B.t.* CRY34/35Ab1 maize lines will be planted in plots of up to 4 rows by 30 feet in length. Each line may be represented both by a segregate without the ICP and a segregate with the ICP. Experimental units (rows) will contain up to 100 plants. Plants will be infested with Western corn rootworm, and/or other corn insects, and/or rely on natural infestations. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Locations

State	Number of locations	State	Number of locations
IA	3	MN	2
IL	4	MO	2
IN	2	NE	2
KS	2	PA	2
MI	2	WI	2

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.00 acre per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings per site: up to two plantings per site.
Planting Dates: 3/1/03 – 8/1/04
Harvest Dates: 6/1/03 – 11/30/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue, whole plant samples, and/or insect samples may be taken up to ten times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed may be returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35Ab1 Maize Efficacy Trial

Objective

Assess the efficacy of genetically modified *B.t.* CRY34/35Ab1 maize lines expressing the binary ICP (insecticidal crystal protein) in controlling Western corn rootworm and/or other Coleopteran insects.

Description

B.t. CRY34/35Ab1 maize lines alone, or crossed with other genetically modified corn lines, will be planted in up to six replications of a randomized design. Each line may be represented both by a segregate without the ICP and a segregate with the ICP. Experimental units (rows) will contain up to 100 plants. Plants will be infested with Western corn rootworm and/or other Coleopteran insects, and/or rely on natural infestations. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of locations	State	Number Of locations
HI	6	NE	2
IA	4	SD	3
IL	2	TN	2
IN	2	TX	2
KS	2	WI	2
MN	2		

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.0 acres per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings per site: Hawaii: up to 4 cycles with up to 5 plantings for a total of 20 plantings per year; all other states – up to 2 plantings per year.
Planting Dates: 3/1/03 - 3/1/04
Harvest Dates: 6/1/03 – 6/1/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue, whole plant samples, and/or insect samples may be taken up to ten times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed may be returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35Ab1 Maize Hybrid Production Plots Trial

Objective

Production of hybrid seed for use in research strip trials and other forms of research testing.

Description

B.t. CRY34/35Ab1 maize lines expressing the binary ICP (insecticidal crystal protein) will be crossed with non-genetically modified or genetically modified lines to make hybrid seed for yield and agronomic testing. In addition, plants may be treated with herbicide, and/or infested with Western corn rootworm or other corn insects, and/or sampled for various laboratory analyses to determine phenotype. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of Locations	State	Number of Locations
HI	6	KS	2
IA	6	MN	2
IL	7	NE	2
IN	3	PR	6

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.00 acres per planting.

Schedule

Maximum number of plantings per cycle: Hawaii and Puerto Rico: up to 4 cycles with up to 5 plantings for a total of 20 plantings per year; all other states – up to 2 plantings per year.
Planting Dates: 3/1/03 - 3/1/04
Harvest Dates: 6/1/03 – 6/1/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue and/or whole plant samples may be taken several times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Plant material remaining after harvest may be may be returned to the plot for soil composting. Seed and plant material produced in these trials that is not saved for further research, analyses, or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35AB1 Maize Regulatory Field Studies Trial

Objective

These studies will provide the test material (plant tissues) needed for regulatory studies with *B.t.* CRY34/35Ab1 maize lines expressing the binary ICP (insecticidal crystal protein).

Description

B.t. CRY34/35Ab1 maize lines will be planted at each location in up to seven replications of a randomized design. *B.t.* CRY34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed. Plants may be treated with herbicide and sampled for various laboratory analyses. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number Of Locations	State	Number Of Locations
IA	4	NE	4
IL	4	SD	2
IN	4	OH	4
KS	4	TX	4
MN	4	WI	4
MO	4		

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.25 acres per planting, not including area required for isolation, if needed.

Schedule

Maximum number of plantings per cycle: up to two plantings per site.
Planting dates: 3/1/03 - 8/1/04
Harvest dates: 6/1/03 – 11/30/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissue and/or whole plant samples may be taken several times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Plant material remaining after harvest may be may be returned to the plot for soil composting. Seed and plant material produced in these trials that is not saved for further research, analyses, or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

B.t. CRY34/35Ab1 Non-Target Trial

Objective

These studies will provide information on the impact of corn rootworm control strategies on non-target arthropod populations commonly found in maize fields.

Description

B.t. CRY34/35Ab1 maize lines expressing the binary ICP (insecticidal crystal protein) will be planted in up to four replications of a randomized design. Experimental units will contain up to 3,000 plants. Populations of non-target arthropods will be monitored up to ten times during the growing season. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test materials will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of locations
IA	2
NE	3

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.25 acres per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings per site: up to two plantings per site.
Planting Dates: 3/1/03 - 8/1/04
Harvest Dates: 6/1/03 – 11/30/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of

transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Sampling methods will include visual observations, sticky trap collections, pitfall traps, and soil samples to capture arthropods located above, on and below ground. All collected arthropods will be taken into a laboratory at Pioneer Hi-Bred for quantification and identification. Soil samples will also be taken into a laboratory at Pioneer Hi-Bred or other laboratory. A Berlese-Tullgren funnel system will be used to extract all arthropods from the sample. Soil will then be returned to the test site.

Plant tissue, and/or whole plant samples, may be taken up to ten times during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed may be returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.

Herbicide Tolerance Study

Objective

Evaluate the effect of herbicide application on *B.t.* CRY34/35Ab1 genetically modified lines expressing the binary ICP (insecticidal crystal protein).

Description

B.t. CRY34/35Ab1 maize lines will be evaluated for resistance to applications of herbicide. Plants may be evaluated for yield, and for effects of herbicide on plant development and agronomic traits. *B.t.* Cry34/35Ab1 maize lines may be crossed with non-genetically modified or genetically modified corn lines or selfed.

Genotypes and Vectors

Test material will consist of dent corn of varying genetic constitution containing:

Vectors	Events
PHP17658	E4497.52.4.3, E4497.71.1.29, E4497.71.1.33

Additional genetically modified and non-genetically modified lines may be included in the total plot acreage.

Locations

State	Number of locations	State	Number of locations
HI	6	MO	2
IA	5	NE	3
IL	7	OH	2
IN	3	PA	2
KS	2	PR	6
MN	4	WI	4

Acreage per Site

Acres of genetically modified lines expressing the binary ICP: up to 1.50 acres per planting, not including area required for isolation, if used.

Schedule

Maximum number of plantings: Hawaii and Puerto Rico - up to 4 cycles with up to 5 plantings for a total of 20 plantings per year; all other states - up to 2 plantings per year.
Planting Dates: 3/1/03 - 3/1/04
Harvest Dates: 6/1/03 – 6/1/04

Border rows

The entire trial site will be surrounded by at least ten rows of an appropriate corn line.

Isolation

One or more of the following methods may be used: (1) Transgenic plants will be located at least 660 feet from receptive silks on corn intended for commercial sale or replanting . (2) Tassels will be kept bagged from prior to anthesis until pollen shed is complete or until tassel is removed from the plant. (3) Temporal shift with monitoring, where the flowering period of transgenic plants will not coincide with presence of receptive silks on plants within 660 feet. (4) Detasseling of test plants prior to onset of anthesis.

Sampling

Plant tissues (e.g., whole plants and grain) will be collected up to two times at each location during the growing season and returned to Pioneer Hi-Bred, Dow AgroSciences, or other laboratories for analyses.

Harvest Procedures

Plots will be harvested by hand or mechanically. If by hand, ears will be placed in cloth or mesh bags of such construction to avoid loss of seed outside of the bags. If machine harvested, seed will be shelled as part of the process. The harvest machine will be thoroughly cleaned prior to exiting from the plot area.

Final Disposition

Any remaining vegetative material will be tilled into the soil at the trial site. Unwanted seed may be returned to the site prior to cultivation for soil composting. Seed and plant material produced in these trials may be used for analyses or saved for further research or future plantings. Unwanted experimental seed will be destroyed.

Volunteer Plants

Volunteer plants will be minimized by growing transgenic material in defined areas in the field and by performing termination procedures outlined above. The use of stakes, other physical markers, or global positioning systems to define the area where the transgenic plants are grown will be used to identify volunteers for later elimination.